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677 South Segoe Rd., Madison, WI 53711 USA

The Influence of Vegetation in Riparian Filterstrips on Coliform Bacteria: II. Survival in Soils

James A. Entry,* Robert K. Hubbard, Janice E. Thies, and Jeffry J. Fuhrmann

ABSTRACT

Survival of total and fecal coliform bacteria was measured in the 0 to 5, 5 to 15, and 15 to 30 cm soil depths at 1, 3, 7, 14, and 90 to 120 d after swine (*Sus scrofa*) wastewater application to riparian filterstrips in southern Georgia during each season of the year. Vegetative treatments evaluated were: (i) 20 m grass–10 m forest, (ii) 10 m grass–20 m forest, and (iii) 10 m grass–20 m maidencane (*Panicum hemitomon* Schult.). During winter, spring, and summer vegetation type in riparian filterstrips did not affect survival of total and fecal coliform bacteria. Total and fecal coliform bacterial numbers were usually higher in the top 0 to 5 cm of soil than in the 5 to 15 and 15 to 30 cm soil depths in all treatments. Total and fecal coliform numbers in the 0 to 5, 5 to 15, and 15 to 30 cm depths declined approximately 10-fold every 7 to 14 d after waste application in all seasons of the year. At 90 to 120 d after waste application, total and fecal coliform numbers in the three soil depths did not differ from riparian filterstrips that did not have animal waste applied. Total coliform bacteria in the 0 to 5, 5 to 15, and 15 to 30 cm soil depths correlated with temperature and moisture in a curvilinear relationship (r^2 = 0.80 , 0.77, and 0.64, respectively). Fecal coliform bacteria in 0 to 5, 6 to 15, and 16 to 30 cm of soil also correlated with temperature and moisture in a curvilinear relationship (r^2 = 0.56 , 0.53, and 0.53, respectively).

THE NUMBER AND SIZE of animal production operations in the USA has been steadily increasing for several decades. Large volumes of animal waste are generated as waste water from urine and animal washing and as semi-solid or solid manure. Because advanced waste water treatments are cost prohibitive for animal waste (USEPA, 1998; Thomas and Law, 1977; Pratt et al., 1977), the economically viable alternative for manure disposal is land application. Land application of animal waste is a means of disposing of the waste and using it as a fertilizer to supply nutrients to crops. Land application of animal waste is a major source of microorganisms pathogenic to humans (Fraser et al., 1998; Howell et al., 1996; 1995; Mawdsley et al., 1995). Liquid-waste discharge into soil follows natural ground water drainage patterns and may contaminate adjoining bodies of surface water. These same bodies of water may be used as sources of drinking water and/or for recreational activities. Therefore, it is critical to keep these lakes and streams free of intestinal pathogens.

The movement of animal waste into surface and4ground water has been increasingly cited as a major factor contributing to movement of nutrients and pathogenic microorganisms into surface and ground water (Mawdsley et al., 1995; Khaeel et al., 1980). The potential for movement of pathogenic bacteria in surface runoff and ground water will in part depend on soil type, climatic and soil conditions, method and amount of manure disposal, and the amount and type of vegetation growing on the site (Entry et al., 2000; Howell et al., 1996; Maswdsley et al., 1995; Canale et al., 1993; Van Donsel et al., 1967). The companion study (Entry et al., 2000) found that land application of swine waste to riparian vegetation increased total and fecal coliform concentrations in soil water and shallow ground water from 10- to 1000-fold. Time and expense in testing for microorganisms pathogenic to humans have led to the use of indicator bacteria of enteric origin to estimate die-off of pathogens in soil and water. It is generally accepted that total and fecal coliform bacteria will be affected in the same manner as human pathogens in soil and water (Fujioka, 1997; Toranzos and McFeters, 1997; Greenberg et al., 1992).

Riparian vegetation acts as a natural filter and removes nutrients and other contaminants through both ground- and surface-water pathways (Hubbard et al., 1998; Snyder et al., 1998; Jordan et al., 1993). However, there has been little research on the effectiveness of forest riparian filterstrips in protecting stream waters from coliform bacteria. Coyne et al. (1998, 1995), Walker et al. (1990) and Young et al. (1980) concluded that 10 m wide grass filterstrips reduced the amount of fecal coliform bacteria in surface runoff from areas where poultry and dairy waste water have been applied by as much as 70%. These authors, however, concluded that 10 m wide grass filterstrips are often inadequate in bringing animal waste water contaminated with fecal coliform bacteria into compliance with water quality standards.

The ability of pathogenic bacteria to survive in the soil environment increases the probability of water contamination after rainfall events. Pathogen survival time in the upper soil varies from 4 to 160 d (Abu-Ashour et al., 1994; Sjogren, 1994). Survival of pathogenic bacteria first reflects the organism's ability to respond to nonparasitic and adverse environmental conditions. Obligate parasites usually only live a few minutes outside the host, but many pathogenic organisms can live in the soil for months (Sorber and Moore, 1987). Several factors influence the survival of pathogens in soil after waste materials are applied. Soil moisture seems to be the most important of these factors (Sjogren, 1994; Crane and Moore, 1986). Survival of bacteria that are pathogenic to humans in soil increases when the soil is moist. Soil temperature also exerts a major influence on the survival of pathogenic bacteria. Extremely hot (>28°C)

James A. Entry, USDA-ARS, Northwest Irrigation and Soils Research Lab., 3793 North 3600 East, Kimberly, ID 83443. Robert K. Hubbard, USDA-ARS, Southeast Watershed Research Lab., P.O. Box 946, Tifton, GA 31793-0748. Janice E. Thies, Cornell Univ., Dep. of Crop and Soil Science, 722 Bradfield Hall, Ithaca, NY 14853. Jeffry J. Fuhrmann, Dep. of Plant and Soil Sciences, Univ. of Delaware, Newark, DE 19717-1303. Received 14 June 1999. *Corresponding author (jentry@kimberly.ars.pn.usbr.gov).

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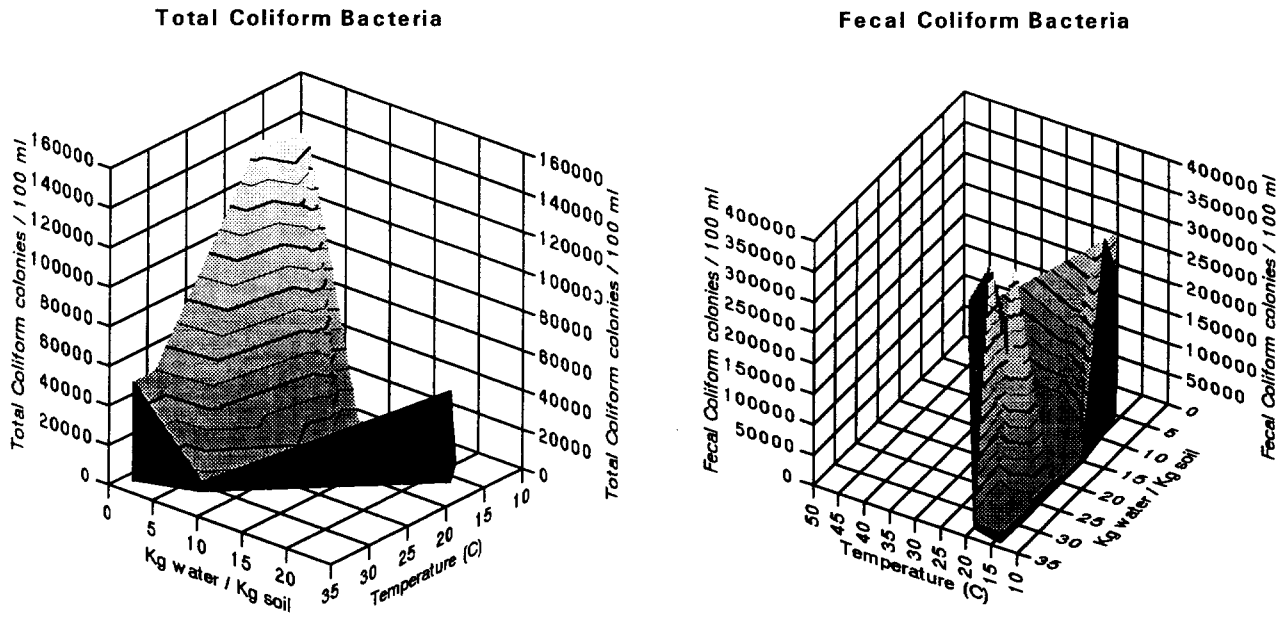

Fig. 1. Numbers of total (left graph) and fecal (right graph) coliform bacteria in the 0 to 5 cm layer of riparian soils. Numbers of total coliform bacteria in soil were explained by the following polynomial regression with soil temperature (ST) and soil moisture (SM). Numbers of total coliform bacteria = 2.4188 + 0.386 (ST) − 0.0098 (ST)^2 + 0.0000014 (ST)^3 − 0.108 (SM) + 0.0031 (SM)^2 + 0.000007 (SM)^3, r^2 = 0.80 (p < 0.0001). Numbers of fecal coliform bacteria = 4.8745 + 0.1729 (ST) − 0.0055 (ST)^2 + 0.0000085 (ST)^3 − 0.1974 (SM) + 0.0050 (SM)^2 + 0.000001 (SM)^3, r^2 = 0.56 (p < 0.0001).

soil temperatures combined with drying will effectively decrease survival rates (Sjogren, 1994; Reddy et al., 1981). The objective of this study was to determine the survival of total and fecal coliform bacteria in soil growing three different types of riparian vegetation in different climatic conditions.

MATERIALS AND METHODS

Site Description

Plots were located at the Animal Science Research Farm at the Coastal Plain Experiment Station in Tifton, Georgia. The site included a grassed area that had formerly been the lowest end of a pasture for beef cattle (*Bos taurus*), an adjacent downslope riparian forest with slash pine (*Pinus elliottii* Engelm.), and accompanying underlying shrubby vegetation (Hubbard et al., 1998). The soil of the grassed area was Tifton loamy sand (fine-loamy, kaolinitic, thermic Plinthic Kandiudults) while the riparian forest area was an Alapaha loamy sand (loamy, silicious, thermic Arenic Plinthic Paleaquults) or an intergrade between it and Tifton loamy sand. These soils are underlain with plinthite and Miocene age materials of very low permeability. In the plinthic soils of the Tifton Upland, 99% of infiltrating water moves downslope as shallow lateral flow (Hubbard and Sheridan, 1983). The slope at the site ranged from 1.5 to 2.0%.

Three different vegetation types were used for the study (Fig. 1 and 2; Entry et al., 2000). Vegetative treatments were (i) 20 m grass buffer draining into 10 m existing forest riparian zone vegetation, (ii) 10 m grass buffer draining into 20 m existing forest riparian zone vegetation, and (iii) 10 m grass buffer draining into 20 m maidencane. Maidencane is a species recommended for constructed wetlands. The purpose of maidencane was to see if wetland plant species other than trees and understory vegetation would be effective in reducing survival of total and fecal coliform bacteria compared with grass and forest vegetation. Maidencane was planted as rhizomes during the summer of 1993. Three or four cuttings of the grassed zone were made each summer and the biomass was completely removed from the plots. Two cuttings (at approximately 30 cm height) were made of the maidencane during the summers. Maidencane is used as forage in Florida, so cutting and removal of the maidencane biomass was used to simulate potential cutting of this plant for hay by animal producers.

Coastal bermuda grass [*Cynodon dactylon* (L.) Pers; cv.Tifton 78] was planted in the grassed portion of the plots. During the fall of 1993 Georgia 5 fescue (*Festuca arundinacea* Schreb.), a heat-tolerant tall fescue, was planted as a perennial winter cover in the grassed portion of the plots. During the winter of 1995–1998, crimson clover (*Trifolium incarnatum* L.) was seeded on the plots, since the fescue had not performed well in terms of cover during the previous winter (1994–1995). The forested part of the plots had slash pine that were 8 yr old and approximately 10 m tall by the end of the study in 1999. In this study grass was cut to a 10 cm height prior to each application of swine waste, while maidencane grew to a height of approximately 30 cm in spring and 2 m in summer.

Plot Design

The study was arranged in a completely randomized factorial design and consisting of three vegetation types and four different seasons having four distinct climatic conditions. Numbers of total and fecal coliform bacteria were sampled in winter (wet–cool period; 14–28 January), spring (warm–cool; 16–30 March), summer (hot–dry; 6–21 July), and autumn (dry–cool; 11–25 November) 1998. Forest and grass filterstrip soils that did not have animal waste applied, or were grazed in the same area and soil type as the vegetative filterstrips that received swine waste, were sampled as controls. The overland flow–riparian buffer plots were each 4 m wide and 30 m long, and were positioned on the landscape according to contour, so that flow of the wastewater downslope would be as uniform as possible (Entry et al., 2000). The sides of each plot were

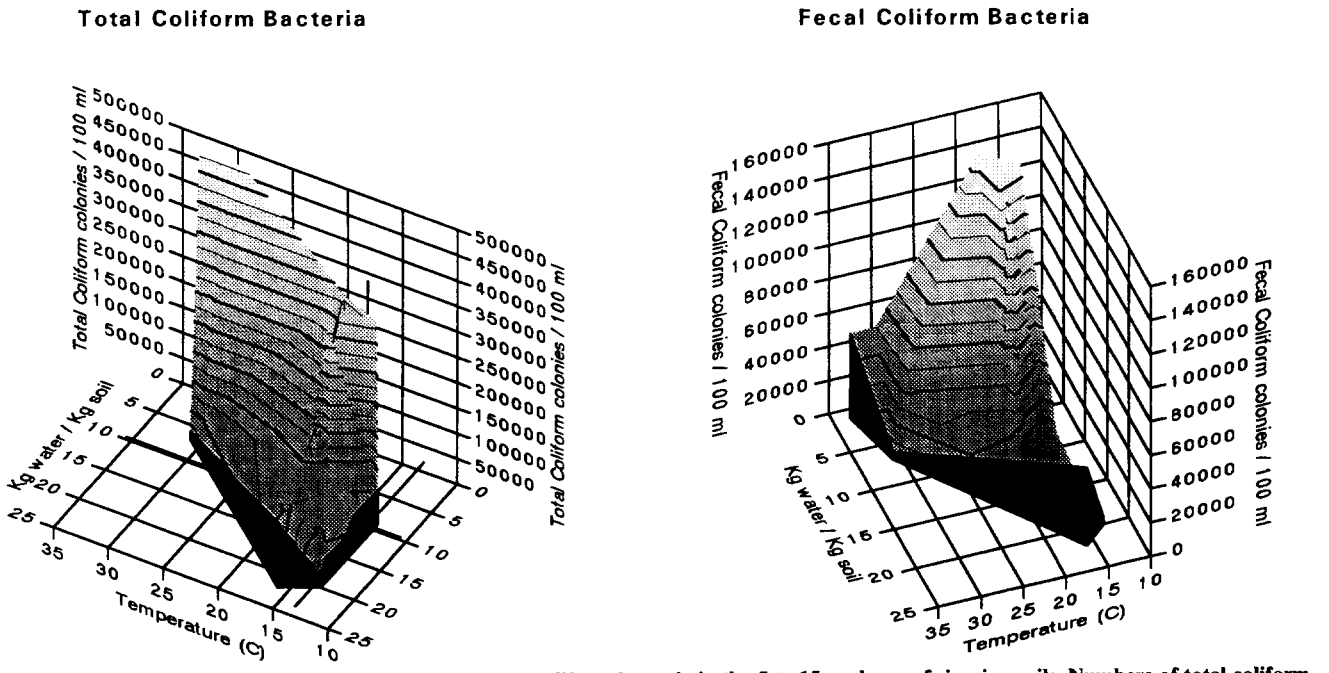

Fig. 2. Numbers of total (left graph) and fecal (right graph) coliform bacteria in the 5 to 15 cm layer of riparian soils. Numbers of total coliform bacteria in soil were explained by the following polynomial regression with soil temperature (ST) and soil moisture (SM). Numbers of total coliform bacteria = 1.5614 + 0.6346 (ST) − 0.02154 $(ST)^2$ − 0.0000058 $(ST)^3$ − 0.2862 (SM) + 0.01339 $(SM)^2$ + 0.000008 $(SM)^3$, r^2 = 0.77 ($p < 0.0001$). Numbers of fecal coliform bacteria = 6.7534 + 0.112 (ST) − 0.0017$(ST)^2$ + 0.0000026 $(ST)^3$ − 0.0422 (SM), − 0.0022 $(SM)^2$ + 0.0000041 $(SM)^3$, r^2 = 0.53 ($p < 0.0001$).

bounded with plastic borders that extended 15 cm above ground and 15 cm below ground. An earthen berm at the top of each plot prevented surface runoff entering from upslope of the plot. At the top of each plot a gated pipe made of plastic was used to apply wastewater.

The individual plots were positioned on the landscape to minimize cross-contamination of the shallow ground water and vegetative treatments (Entry et al., 2000). Wastewater flowed from an individual tank above each plot into the gated pipe and then downslope. The pipe gates were spaced 46 cm apart and were adjusted on each plot so that overland flow would begin downslope movement as uniformly as possible. Depending on soil moisture and vegetative conditions, wastewater flowed over one-half to two-thirds of each plot during application. Wastewater was not applied during rainstorms or if it appeared that rainfall was imminent. During the wet winter and spring months when the soil was nearly saturated, the wastewater was applied slowly, to minimize any potential for the waste to exit the plots via overflow of the plastic borders. At other times the wastewater was applied as quickly as the tanks would drain completely, resulting in application times of approximately 10 min.

Wastewater

The study was implemented using swine lagoon wastewater from the treatment–storage system at the University of Georgia Coastal Plain Experiment Station swine unit at Tifton, Georgia. The unit maintained an inventory of 350 to 550 head of swine during the course of the study. The lagoon system consisted of three lagoons, in series. The primary lagoon (12 × 46 m) discharged into a secondary lagoon, with a (15 × 31 m) from which the liquid was pumped back into the barns (1800 L min^{-1}) for flushing waste. The secondary lagoon discharged into a holding lagoon (18 × 37 m) that was used as the source of the wastewater for the study. Features of the lagoon system have been further described (Hubbard et al. 1998; Newton and Haydon, 1985). Eight hours prior to application, liquid waste was pumped approximately 760 m from the holding lagoon into 2 m diameter × 1.5 m high plastic holding tanks located at the upper end of each at each plot (Entry et al., 2000). Samples of wastewater application consisted of 2570 L, from two tanks, per plot as one application each season, which is a typical amount of animal wastewater applied to a site to meet crop N demands (Hubbard et al. 1998).

Total coliform numbers in source wastewater ranged from 4.92 × 10^6 to 10.5 × 10^5 colonies 100 mL $water^{-1}$. Numbers of fecal coliform bacteria in source wastewater ranged from 15.8 × 10^5 to 7.0 × 10^5 colonies 100 mL $water^{-1}$. Analyses of wastewater samples collected weekly during the study showed average N concentration of 160 mg L^{-1}, with most of the N in the NH_4^+–N form. Nitrate concentrations in the wastewater ranged from less than 1 mg L^{-1} to 20 mg L^{-1} with a mean of 3 mg L^{-1}.

Experimental Design

The study was arranged in a completely random factorial design consisting of filterstrips with three vegetation types, climatic periods (season), distance from inflow source, and depth of soil water and ground water (Kirk, 1982). Each treatment was replicated three times. Wastewater was applied as a single pulse in winter, spring, summer, and autumn having four distinct climatic conditions (described below). Soil was collected at two distances from the wastewater inflow source. Survival of total and fecal coliform bacteria in each combination of vegetation type and climate was determined at 7.0 and 14.5 m from the inflow point at the 0 to 5, 5 to 15, and 15 to 30 cm soil depths at 1, 3, 7, 14, and 90 to 120 d after application of wastewater.

Total and fecal coliform bacteria were sampled in winter (wet–cool period; 14–28 January), spring (warm–wet; 16–30 March), summer (hot–dry; 6–21 July), and autumn (warm–dry; 11–25 November) 1998. In winter soil temperature ranged

Table 1. Survival of total coliform bacteria in soil growing three different types of riparian vegetation after application of swine waste water.†

		Winter					Spring				
		Day					Day				
Vegetation	Depth	−1 (90)	1	3	7	14	−1 (63)	1	3	7	14
	cm	Total coliform bacteria									
		bacteria/g^{-1} soil									
Control/grass filterstrip‡	0–5	13.3 × 10^{2}c					21.0 × 10^{4}b				
	5–15	13.3 × 10^{2}c					36.0 × 10^{3}c				
	15–30	10.0 × 10^{2}c					14.0 × 10^{3}c				
Control/forest filterstrip‡	0–5	23.3 × 10^{2}c					13.7 × 10^{3}c				
	5–15	6.6 × 10^{2}c					50.0 × 10^{2}cd				
	15–30	3.3 × 10^{2}c					30.0 × 10^{2}d				
Waste source (Inflow)§	0		10.5 × 10^{5}a					49.2 × 10^{6}a			
20 m grass/ 10 m forest	0–5	96.6 × 10^{3}b	14.1 × 10^{5}a	47.3 × 10^{4}b	43.7 × 10^{4}b	93.3 × 10^{3}b	96.7 × 10^{2}cd	30.0 × 10^{2}c	28.1 × 10^{5}ab	61.8 × 10^{4}b	23.3 × 10^{4}b
	5–15	15.3 × 10^{3}c	30.5 × 10^{4}b	21.5 × 10^{3}b	56.3 × 10^{4}b	66.6 × 10^{3}b	15.3 × 10^{3}c	43.3 × 10^{2}c	10.9 × 10^{5}b	24.5 × 10^{4}b	75.0 × 10^{3}b
	15–30	23.3 × 10^{2}c	85.3 × 10^{3}c	13.6 × 10^{3}b	39.1 × 10^{4}b	73.3 × 10^{3}b	23.3 × 10^{2}d	33.3 × 10^{2}c	11.1 × 10^{5}ab	40.0 × 10^{3}b	61.6 × 10^{3}bc
10 m grass/ 20 m forest	0–5	26.1 × 10^{3}c	14.0 × 10^{4}b	82.0 × 10^{3}b	18.4 × 10^{4}b	54.3 × 10^{3}b	26.1 × 10^{3}c	50.0 × 10^{2}c	42.3 × 10^{4}b	12.0 × 10^{4}b	23.1 × 10^{3}c
	5–15	56.6 × 10^{2}c	20.0 × 10^{4}b	15.8 × 10^{3}b	72.9 × 10^{4}b	20.5 × 10^{3}b	56.6 × 10^{3}c	36.2 × 10^{2}c	96.8 × 10^{4}ab	42.5 × 10^{4}b	41.6 × 10^{3}bc
	15–30	10.0 × 10^{2}c	11.8 × 10^{4}b	83.3 × 10^{2}b	24.1 × 10^{4}b	13.8 × 10^{3}b	10.0 × 10^{2}d	13.3 × 10^{2}c	17.3 × 10^{4}b	33.3 × 10^{3}bc	22.3 × 10^{3}c
10 m grass/ 20 m maidencane	0–5	10.3 × 10^{3}c	88.5 × 10^{4}b	18.0 × 10^{4}b	84.6 × 10^{4}b	28.0 × 10^{3}c	10.3 × 10^{3}c	13.3 × 10^{2}c	60.4 × 10^{4}b	37.5 × 10^{4}b	20.0 × 10^{4}b
	5–15	18.8 × 10^{3}c	13.6 × 10^{4}b	51.6 × 10^{2}bc	20.7 × 10^{4}b	31.7 × 10^{3}c	18.3 × 10^{3}c	66.6 × 10^{2}c	12.6 × 10^{5}ab	14.0 × 10^{4}b	44.0 × 10^{3}bc
	15–30	21.3 × 10^{3}c	91.1 × 10^{3}c	26.6 × 10^{2}bc	74.5 × 10^{3}b	78.3 × 10^{3}b	21.3 × 10^{3}c	18.3 × 10^{2}c	22.0 × 10^{4}b	75.0 × 10^{3}b	25.0 × 10^{3}c

Continued.

Table 1. Continued.†

Vegetation	Depth	Summer					Autumn					
		Day					Day					
		−1 (130)	1	3	7	14	−1 (120)	1	3	7	14	98
	cm	Total coliform bacteria										
		bacteria/g^{-1} soil										
Control/grass filterstrip‡	0–5	13.6 × 10^{3}c					2.9 × 10^{4}c					
	5–15	50.0 × 10^{2}d					3.6 × 10^{4}c					
	15–30	30.0 × 10^{2}d					1.7 × 10^{4}c					
Control/forest filterstrip‡	0–5	21.0 × 10^{4}d					3.2 × 10^{4}c					
	5–15	36.0 × 10^{3}c					9.6 × 10^{3}cd					
	15–30	14.0 × 10^{3}c					11.0 × 10^{3}cd					
Waste source (Inflow)§	0		11.1 × 10^{6}a					10.4 × 10^{6}a				
20 m grass/ 10 m forest	0–5	23.0 × 10^{3}c	96.9 × 10^{4}b	12.4 × 10^{4}b	14.0 × 10^{4}b	79.8 × 10^{4}b	29.6 × 10^{3}c	8.5 × 10^{5}b	8.5 × 10^{5}b	64.7 × 10^{4}c	74.1 × 10^{4}c	44.1 × 10^{4}c
	5–15	31.6 × 10^{3}c	116.7 × 10^{4}b	10.6 × 10^{4}b	13.1 × 10^{4}b	54.6 × 10^{3}c	3.5 × 10^{3}d	3.2 × 10^{6}b	4.3 × 10^{5}b	44.7 × 10^{4}c	33.7 × 10^{4}c	17.6 × 10^{4}c
	15–30	33.3 × 10^{2}d	111.6 × 10^{4}b	11.8 × 10^{4}b	10.3 × 10^{4}b	30.8 × 10^{3}c	17.6 × 10^{3}d	2.0 × 10^{5}b	17.1 × 10^{4}c	64.8 × 10^{3}cd	16.3 × 10^{3}d	10.3 × 10^{3}d
10 m grass/ 20 m forest	0–5	23.5 × 10^{3}c	96.6 × 10^{4}b	103.3 × 10^{4}b	110.6 × 10^{4}b	62.0 × 10^{4}b	9.6 × 10^{3}d	17.5 × 10^{4}c	16.3 × 10^{4}c	28.6 × 10^{4}c	46.8 × 10^{3}cd	42.6 × 10^{4}c
	5–15	102.0 × 10^{2}d	50.0 × 10^{3}bc	44.6 × 10^{4}b	101.0 × 10^{3}b	16.2 × 10^{3}c	12.1 × 10^{3}d	18.0 × 10^{4}c	15.0 × 10^{4}c	13.4 × 10^{4}c	12.4 × 10^{4}c	17.8 × 10^{3}c
	15–30	16.7 × 10^{2}d	73.3 × 10^{3}bc	14.4 × 10^{4}b	74.0 × 10^{3}c	32.0 × 10^{3}c	11.0 × 10^{3}d	19.0 × 10^{4}c	13.9 × 10^{4}c	28.1 × 10^{3}d	17.8 × 10^{3}d	52.6 × 10^{3}d
10 m grass/ 20 m maidencane	0–5	100.0 × 10^{3}c	110.0 × 10^{4}b	40.2 × 10^{4}b	17.8 × 10^{4}b	82.0 × 10^{3}bc	2.1 × 10^{3}d	19.8 × 10^{4}c	34.4 × 10^{4}c	36.9 × 10^{4}c	35.0 × 10^{4}c	60.0 × 10^{3}d
	5–15	120.0 × 10^{3}c	76.6 × 10^{4}bc	80.0 × 10^{4}b	12.8 × 10^{4}b	31.3 × 10^{3}c	6.1 × 10^{3}d	21.3 × 10^{4}c	25.6 × 10^{4}c	21.7 × 10^{4}c	12.3 × 10^{4}c	21.5 × 10^{4}c
	15–30	125.0 × 10^{3}c	50.0 × 10^{3}c	57.6 × 10^{3}c	11.2 × 10^{4}b	36.6 × 10^{3}c	6.5 × 10^{3}d	14.0 × 10^{4}c	15.1 × 10^{4}c	48.3 × 10^{3}d	15.5 × 10^{3}d	15.7 × 10^{3}d

† In each season, in each column and row, values followed by the same letter are not significantly different as determined by the least square means test ($P \leq 0.05$), $n = 16$. Tables can be read both vertically and horizontally beginning with the waste source value which was sampled immediately prior to waste application.

‡ Soils were sampled at Gibbs Farm (Lowrance et al., 1998) where no animal waste has been applied.

§ Waste was applied at Day 0; on Day −1, soil was sampled prior to application of swine waste and 63–130 d after the last application of swine waste on these plots.

Table 2. Survival of fecal coliform bacteria in soil growing three different types of riparian vegetation after application of swine waste water.†

		Winter					Spring				
		Day					Day				
Vegetation	Depth	−1 (90)	1	3	7	14	−1 (63)	1	3	7	14
	cm	Fecal coliform bacteria, bacteria/g^{-1} soil									
Control/grass‡	0–5	13.3 × 10^{2}c					50.0 × 10^{1}				
	5–15	10.3 × 10^{2}c					0.0				
	15–30	0.0 × 10^{0}f					0.0				
Control/forest‡	0–5	23.3 × 10^{2}c					33.3 × 10^{0}				
	5–15	0.0 × 10^{0}f					0.0				
	15–30	0.0 × 10^{0}f					0.0				
Waste source (Inflow)§	0		11.6 × 10^{5}a					15.8 × 10^{4}a			
20 m grass/ 10 m forest	0–5	13.3 × 10^{2}c	10.3 × 10^{4}b	41.0 × 10^{3}b	69.8 × 10^{2}bc	14.2 × 10^{2}c	23.3 × 10^{3}b	11.6 × 10^{3}b	85.0 × 10^{2}bc	81.1 × 10^{1}cd	61.6 × 10^{1}e
	5–15	13.3 × 10^{2}c	76.6 × 10^{3}b	47.6 × 10^{2}bc	55.8 × 10^{2}bc	57.6 × 10^{1}cd	50.0 × 10^{2}bc	10.6 × 10^{3}b	46.0 × 10^{2}bc	46.8 × 10^{1}cd	16.6 × 10^{1}e
	15–30	0.0f	68.3 × 10^{3}b	57.9 × 10^{2}bc	28.3 × 10^{2}c	46.8 × 10^{1}cd	62.1 × 10^{2}bc	20.3 × 10^{2}c	33.3 × 10^{1}d	28.3 × 10^{1}d	33.3 × 10^{1}e
10 m grass/ 20 m forest	0–5	0.0f	26.8 × 10^{3}b	33.8 × 10^{2}bc	12.8 × 10^{2}c	67.2 × 10^{1}cd	8.9 × 10^{3}b	50.0 × 10^{2}b	12.6 × 10^{2}c	50.0 × 10^{1}e	9.8 × 10^{1}e
	5–15	0.0f	65.0 × 10^{3}b	84.6 × 10^{2}bc	21.2 × 10^{2}c	42.1 × 10^{1}d	10.6 × 10^{3}b	63.8 × 10^{2}c	17.9 × 10^{2}c	83.3 × 10^{1}e	20.6 × 10^{1}e
	15–30	0.0f	90.0 × 10^{3}b	56.7 × 10^{2}bc	10.0 × 10^{2}c	88.6 × 10^{1}cd	52.7 × 10^{2}bc	28.2 × 10^{2}c	16.7 × 10^{2}c	21.2 × 10^{1}e	16.7 × 10^{1}e
10 m grass/ 20 m maidencane	0–5	0.0f	44.8 × 10^{3}b	10.8 × 10^{3}c	50.0 × 10^{2}bc	13.6 × 10^{2}c	33.3 × 10^{3}b	17.6 × 10^{3}b	29.5 × 10^{2}bc	50.0 × 10^{1}cd	50.0 × 10^{1}e
	5–15	0.0f	50.8 × 10^{3}b	57.8 × 10^{2}bc	28.3 × 10^{2}c	28.3 × 10^{1}d	12.2 × 10^{3}b	33.3 × 10^{2}b	18.3 × 10^{2}c	28.3 × 10^{1}d	20.0 × 10^{1}e
	15–30	0.0f	41.6 × 10^{3}b	87.5 × 10^{2}bc	23.3 × 10^{2}c	33.3 × 10^{1}d	5.7 × 10^{3}b	15.3 × 10^{2}c	11.6 × 10^{2}c	23.3 × 10^{1}d	10.0 × 10^{1}e

Continued.

Table 2. Continued.†

Vegetation	Depth	Summer Day −1 (130)	Summer Day 1	Summer Day 3	Summer Day 7	Summer Day 14	Autumn Day −1 (120)	Autumn Day 1	Autumn Day 3	Autumn Day 7	Autumn Day 14	Autumn Day 98
	cm	Fecal coliform bacteria — bacteria/g^{-1} soil										
Control/grass‡	0–5	33.0 × 10^{1}e					6.3 × 10^{3}d					
	5–15	00.0f					2.1 × 10^{3}d					
	15–30	00.0f					2.7 × 10^{3}d					
Control/forest‡	0–5	50.0 × 10^{1}e					4.0 × 10^{3}d					
	5–15	0.0f					3.3 × 10^{3}d					
	15–30	0.0f					0.0 × 10^{0}f					
Waste source (Inflow)§	0		7.70 × 10^{5}a					2.9 × 10^{6}a				
20 m grass/ 10 m forest	0–5	0.0f	71.6 × 10^{3}b	12.6 × 10^{4}b	22.5 × 10^{3}b	70.8 × 10^{3}b	6.3 × 10^{2}e	32.0 × 10^{5}b	35.0 × 10^{4}c	41.1 × 10^{4}c	27.5 × 10^{4}c	6.7 × 10^{4}c
	5–15	0.0f	41.0 × 10^{3}b	60.8 × 10^{3}b	18.1 × 10^{3}b	49.5 × 10^{2}bc	2.1 × 10^{2}e	14.1 × 10^{5}b	73.0 × 10^{4}c	15.5 × 10^{4}c	14.7 × 10^{4}c	12.8 × 10d
	15–30	0.0f	77.6 × 10^{3}b	31.5 × 10^{2}b	32.5 × 10^{2}c	13.6 × 10^{2}c	2.6 × 10^{2}e	12.3 × 10^{5}b	3.2 × 10^{4}c	10.8 × 10^{3}d	20.3 × 10^{3}d	0.0f
10 m grass/ 20 m forest	0–5	0.0f	21.1 × 10^{3}b	11.3 × 10^{3}b	34.0 × 10^{3}b	29.3 × 10^{3}b	3.3 × 10^{3}e	16.3 × 10^{4}c	20.4 × 10^{4}c	19.1 × 10^{3}d	31.7 × 10^{3}c	8.9 × 10^{4}c
	5–15	0.0f	34.3 × 10^{2}b	10.0 × 10^{3}b	70.0 × 10^{1}c	55.3 × 10^{1}c	3.9 × 10^{3}e	18.4 × 10^{4}c	9.8 × 10^{4}c	2.9 × 10^{3}d	46.1 × 10^{3}c	3.3 × 10^{3}d
	15–30	0.0f	23.3 × 10^{2}b	38.0 × 10^{3}b	33.0 × 10^{2}bc	17.3 × 10^{1}d	0.0 × 10^{0}f	8.2 × 10^{4}c	4.5 × 10^{4}c	5.1 × 10^{3}d	11.6 × 10^{2}e	10.0 × 10^{2}e
10 m grass/ 20 m maidencane	0–5	0.0f	125.0 × 10^{3}b	15.6 × 10^{3}b	37.0 × 10^{3}b	10.2 × 10^{2}b	12.1 × 10^{2}e	17.1 × 10^{4}c	17.3 × 10^{4}c	26.4 × 10^{4}c	23.2 × 10^{4}c	32.8 × 10^{3}c
	5–15	0.0f	33.3 × 10^{3}b	72.0 × 10^{2}c	89.3 × 10^{2}bc	93.3 × 10^{2}bc	3.8 × 10^{2}e	73.8 × 10^{3}cd	37.0 × 10^{3}cd	49.8 × 10^{3}cd	42.4 × 10^{3}c	13.3 × 10^{3}d
	15–30	0.0f	21.0 × 10^{3}b	35.2 × 10^{2}c	91.3 × 10^{1}c	0.0 × 10^{0}f	13.5 × 10^{2}e	5.0 × 10^{3}d	19.5 × 10^{3}d	4.1 × 10^{3}d	8.3 × 10^{2}e	5.5 × 10^{3}d

† In each season, in each column and row, values followed by the same letter are not significantly different as determined by the least square means test ($P \leq 0.05$), $n = 16$. Tables can be read both vertically and horizontally beginning with the waste source value which was sampled immediately prior to waste application.

‡ Soils were sampled at Gibbs Farm (Lowrance et al., 1998) where no animal waste has been applied.

§ Waste was applied at Day 0; on Day −1, soil was sampled prior to application of swine waste and 63–130 d after the last application of swine waste on these plots.

from 13 to 14°C and soil moisture ranged from 1.8 to 2.8 g kg soil^{-1}, in spring soil temperature ranged from 17 to 22°C and soil moisture ranged from 1.5 to 3.3 g kg soil^{-1}, in summer soil temperature ranged from 30 to 45°C and soil moisture ranged from 0.1 to 0.7 g kg soil^{-1}, and in autumn soil temperature ranged from 13 to 18°C and soil moisture ranged from 0.3 to 1.1 g kg soil^{-1}. Soil temperature varied from 13°C in winter to 22°C in spring. Soil temperature did not vary appreciably among filterstrip vegetation types in any season.

Sample Collection

We collected two sample cores each at 7.0 and 14.5 m downslope from the waste application point at the top of each plot. Each core was divided into three depths, 0 to 5, 6 to 15, and 16 to 30 cm, placed in sterile plastic bags and transported to and processed (within 6 hr) at the Agricultural Research Service Southeast Watershed Research Laboratory in Tifton, GA.

Sample Analysis

Samples were stored at ambient temperatures and incubation began within 6 hr of collection. A 10 g subsample was placed in 90 mL of dilute phosphate buffer (Greenberg et al., 1992) in a 160 mL container and shaken for 30 min on a Eberbach (Ann Arbor, MI) shaker at 60 cycles min^{-1}. Containers were then removed and further diluted to 10^{-3} to 10^{-5} with phosphate buffer for filtration through membranes and placement on appropriate microbiological medium. Total and fecal coliform bacteria were analyzed with the membrane filter technique (Greenberg et al., 1992). Preliminary samples of soil extracts in each filterstrip were analyzed to determine each dilution before bacteria were counted. One gram of soil was diluted in phosphate buffer (Greenberg et al., 1992) in a series of three to five. A 100-mL sample of the final dilution of each sample was vacuum-filtered through a sterile 0.45-μm filter and placed on m-Endo LES medium to determine total coliform bacteria or FC medium to determine fecal coliform bacteria. Total and fecal coliform bacteria were incubated at 39.5°C ± 0.02 and 44.5°C ± 0.02 respectively for 24 h. Three colony types of both total and fecal coliform bacteria from each sample date were identified by fatty acid analysis using the AEROBE library of the Microbial Identification System (Newark, DE).

Statistical Analyses

All dependent variables were tested for normal distribution. Data were then analyzed by means of analysis of variance procedures (ANOVA) for a completely random design with Statistical Analysis Systems (SAS, 1996). Numbers of total and fecal coliform bacteria were transformed using logarithms to achieve normal distributions. Statistical comparisons were made of total and fecal coliform bacteria by vegetation type × season × time since application at each soil depth. Residuals were equally distributed with constant variances. Differences reported throughout are significant at $p \leq 0.05$, as determined by the protected Least Squares Means (LSD) test (Snedechor and Cochran, 1980; Kirk, 1982). Correlations were calculated using soil temperature and moisture as dependent (x) variables and total or fecal coliform bacteria as independent (y) variables. Total and fecal coliform bacteria are reported in untransformed numbers.

RESULTS

Except for temperature in the top 0 to 5 cm of soil in the summer, temperature and moisture conditions in the top 0 to 5 and 5 to 30 cm of soil did not vary more than 3°C among filterstrip vegetation types during any season. The general linear models procedure indicated that there was no significant difference in total and fecal coliform bacterial numbers sampled in soil at 7.0 and 14.5 m at 0 to 5, 5 to 15 and 15 to 30 cm depths regardless of vegetative treatment or season of year. Therefore, total and fecal coliform bacterial numbers will be discussed with regard to soil depth × vegetative treatment × season of year (Kirk, 1982; Snedecor and Cochran, 1980).

During autumn, the 20 m grass–10 m forest filterstrip had generally higher total and fecal coliform numbers in the 0 to 5 and 5 to 15 cm soil depths than the 10 m grass–20 m forest and 10 m grass–20 m maidencane filterstrips (Tables 1 and 2). Total and fecal coliform bacteria were 10-fold higher in the waste source prior to application than in the soil 24 hr after waste was applied. Total and fecal coliform bacterial numbers were usually higher in the top 0 to 5 cm of soil than in the 5 to 15 and 15 to 30 cm of soil in all treatments and soils in filterstrips that did not have waste applied (controls), although the difference was generally not statistically significant. In general, total and fecal coliform bacterial numbers at all three soil depths declined approximately 10-fold every 7 to 14 d for the first 14 d after waste application in all seasons. At 90 to 120 d after waste application, total coliform bacteria in the top 30 cm of soil ranged from 100 to 36 000 g^{-1} soil and fecal coliform bacteria ranged from 0 to 2300 g^{-1} soil and did not differ from riparian filterstrips that did not have animal waste applied (control sites). Mortality of total coliform bacteria at the 0 to 5, 5 to 15 and 15 to 30 cm depths correlated with decreasing moisture and increasing temperature in a curvilinear relationship (r^2 = 0.80, 0.77, and 0.64 respectively) (Fig. 1, 2, and 3). Morality of fecal coliform bacteria at the 0 to 5, 5 to 15, and 15 to 30 cm soil depths also correlated with decreasing moisture and increasing temperature in a curvilinear relationship (r^2 = 0.56 , 0.53, and 0.53 respectively).

DISCUSSION

The contamination of surface and ground water is often dependent on the concentration of pathogenic bacteria present in the soil prior to a rainfall event. Vegetation type in riparian filterstrips usually did not affect survival of total and fecal coliform bacteria in soil. Several factors influence the survival of pathogens in soil after waste materials are applied. We found that decreasing soil moisture with increasing soil temperature substantially decreased survival of total and fecal coliform bacteria in all three soil depths examined. Reddy et al. (1981) found that fecal coliform bacteria die-off follow first order kinetics and that the two most important factors influencing survival were moisture a8d temperature. Soil moisture seems to be the most important of these factors (Sjogren, 1994; Crane and Moore, 1986). Survival of bacteria that are pathogenic to humans in soil increases when the soil is moist. Soil temperature also exerts a major influence on the survival of coliform bacteria. Extremely hot (>28°C) soil temperatures combined with drying will effectively decrease survival rates (Reddey et al., 1981; Sjogren

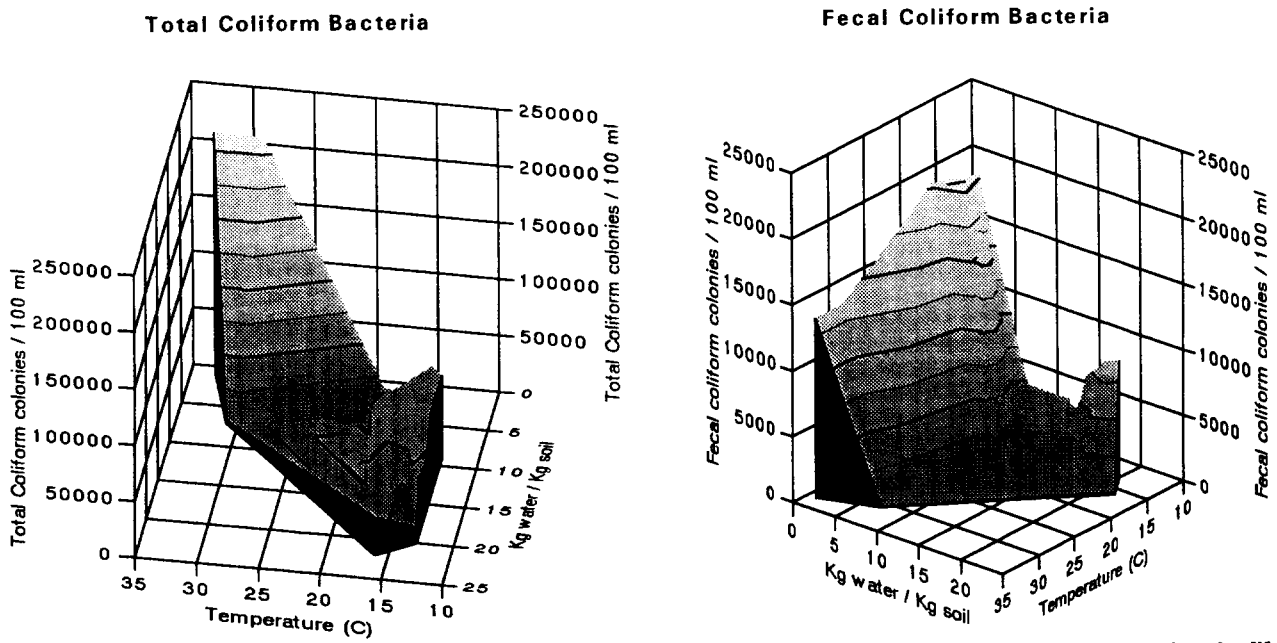

Fig. 3. Numbers of total (left graph) and fecal (right graph) coliform bacteria in the 15 to 30 cm layer of riparian soils. Numbers of total coliform bacteria in soil were explained by the following polynomial regression with soil temperature (ST) and soil moisture (SM). Numbers of total coliform bacteria = 5.4092 + 0.1238 (ST) − 0.0058 $(ST)^2$ + 0.0000022 $(ST)^3$ − 0.4781 (SM) + 0.03438 $(SM)^2$ + 0.0000348 $(SM)^3$, $r^2 = 0.64$ ($p < 0.0001$). Numbers of fecal coliform bacteria = 11.7722 − 0.3667 (ST) + 0.0054 $(ST)^2$ + 0.00000099 $(ST)^3$ − 0.70859 (SM) + 0.0314 $(SM)^2$ + 0.0000187 $(SM)^3$, $r^2 = 0.53$ ($p < 0.0001$).

1994). The amount and availability of nutrients and carbon in soil will also affect pathogen survival. Pathogen survival time in the upper soil varies from 4 to 160 d (Abu-Ashour et al., 1994; Sjogren, 1994). Survival of pathogenic bacteria first reflects the organism's ability to respond to nonparasitic and adverse environmental conditions.

We found that total and fecal coliform bacterial numbers were usually higher in the top 0 to 5 cm of soil than in the 5 to 15 and 15 to 30 cm of soil in all treatments 90 to 120 d after wastewater was applied. Numbers of total and fecal coliform bacteria in soils of all filterstrip treatments after 90 to 120 d after wastewater application were usually not significantly different than riparian filterstrips that did not have waste applied (controls). Grass and forest filterstrips that had no animal waste applied to them had total coliform bacterial numbers ranging from 32 to 36 000 g^{-1} soil and fecal coliform numbers ranging from 0 to 2330 g^{-1} soil. Forest and grass in riparian areas provide habitat for a large variety of warm blooded wildlife who will deposit enteric bacteria in soils and water. Several studies have found significant populations of enteric bacteria in soils and water in areas not influenced by human activity (Buckley et al., 1998; Niemi and Niemi, 1991; Gary et al., 1985). We cannot expect soils or water in areas that are not influenced by anthropogenic activities to be completely free of enteric bacteria. Background concentrations of enteric bacteria in soils and water in areas that are not influenced by human or agricultural operations need to be established to determine if application of human or animal waste is polluting the environment.

Physical and chemical adsorption in soil are the primary mechanisms responsible for the entrapment of pathogenic bacteria. The ability of the soil to filter microorganisms depends largely on its texture and pore space (McMurry et al., 1998; Howell et al., 1996). Many of the pathogenic microorganisms contained in animal and human waste applied to land are filtered out as they move downward through the soil. Gerba et al. (1975) found that 92 to 97% of the *Escherichia coli* are filtered out in the first 4 cm as they move down through the soil. However, both laboratory and field studies have shown that enteric microorganisms can be transported long distances through soil (Huysman and Verstraete, 1993; Van Elsas et al., 1991; Chen, 1988; Smith et al., 1985). The explanation that has been given is the preferential flow of water transporting bacteria through soil macro pores, cracks, and fissures (McMurry et al., 1998; Abu-Ashour et al., 1994). If a significant amount of water is applied to lands where animal waste has been recently applied via rainfall or irrigation, inducing leaching and/or surface runoff, water carrying high concentrations of enteric bacteria will follow natural drainage patterns and may contaminate adjoining bodies of surface water. These same bodies of water may be used for sources of drinking water and/or for recreational activities. Therefore, animal production operations installing vegetative filterstrips may want to pick a vegetation type that will not only establish and grow well in the selected habitat, but also has a high evapotranspiration rate to reduce soil moisture. Animal production operations could also minimize risk of surface and ground water contamination by applying manure or wastewater at times when the soil is expected to be dry for the following 2 to 4 wk.

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